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New technologies: Bioluminescence resonance energy transfer (BRET) for the detection of real time interactions involving G-protein coupled receptors
Pfleger K.D.G.; Eidne K.A.
ADDRESS: Prof. K.A. Eidne, WAIMR, Sir Charles Gairdner Hospital, Hospital Avenue, Perth, WA 6009, Australia
EMAIL: keidne@waimr.uwa.edu.au
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The natural phenomenon of bioluminescence resonance energy transfer (BRET) has become an extremely useful tool for studying protein-protein interactions in the laboratory, including those involving G-protein coupled receptors (GPCRs). The technology involves fusion of donor and acceptor molecules to proteins of interest. Following assessment to ensure correct functionality, co-expression of fusion constructs in live cells enables their interaction to be studied in real time in a quantitative manner. Energy is transferred from the donor to the acceptor when in close proximity, resulting in fluorescence emission at a characteristic wavelength. The energy emitted by the acceptor relative to that emitted by the donor is termed the BRET signal. It is dependent upon the spectral properties, ratio, distance and relative orientation of the donor and acceptor molecules, as well as the strength and stability of the interaction between the proteins of interest. The ability to study interactions in live mammalian cells circumvents many of the problems associated with techniques such as co-immunoprecipitation and yeast two-hybrid screening. Furthermore, the high sensitivity of BRET enables the study of proteins at physiological concentrations, a significant advantage over techniques that require high levels of protein expression. BRET technology has already made a substantial contribution to our understanding of GPCRs and protein-protein interactions, in particular by providing strong evidence that GPCRs homo- and hetero-oligomerize. New BRET detection systems and the potential for novel high throughput screening applications means that BRET promises to play an important role in future research and drug discovery.

DESCRIPTORS:
Bioluminescence resonance energy transfer; BRET; BRETSUP2; Protein-protein interaction; G-protein coupled receptor; GPCR

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